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*Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.*

(54) Title: IMPROVEMENTS RELATING TO DRUG DELIVERY SYSTEMS

(57) Abstract

A two component system designed for use in association with one another comprises (i) a first component that is an antibody fragment capable of binding with a tumour associated antigen, the antibody fragment being bound to an enzyme capable of converting a cytotoxic pro-drug into a cytotoxic drug, (ii) a second component that is a cytotoxic pro-drug convertible under the influence of the enzyme to the cytotoxic drug. This system can be used to control neoplastic cell growth and is designed to improve localisation of the cytotoxic drug. The system utilises benzoic acid nitrogen mustard glutamides convertible to the nitrogen mustard under the influence of carboxypeptidases.

WE CLAIM:

1. A two component system designed for use in association with one another comprising

(i) a first component that is an antibody fragment capable of binding with a tumour associated antigen, the antibody fragment being conjugated to an enzyme capable of converting a cytotoxic pro-drug into an cytotoxic drug.

(ii) a second component that is a cytotoxic pro-drug convertible under the influence of the enzyme to the cytotoxic drug.

2. A system according to claim 1 wherein the antibody is a monoclonal antibody.

3. A system according to claim 1 or 2 wherein the antibody is of the IgG class.

4. A system according to any one of the preceding claims wherein the antibody fragment is a $F(ab')_2$ fragment.

5. A system according to any one of the preceding claims wherein the enzyme is of non-mammalian origin.

6. A system according to any one of the preceding claims wherein the enzyme is a carboxypeptidase.

7. A system according to any one of the preceding claims wherein the pro-drug is an amide obtainable by reaction of a disubstituted amino benzoic acid nitrogen mustard and an amino acid.

8. A system according to claim 7 wherein the amino

acid is a D-amino acid.

10. A system according to any one of claims 7 to 9 wherein the nitrogen mustard is p-bis(2-chloroethyl)amino-benzoic acid.

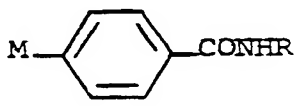
11. A system according to any one of claims 7 to 9 wherein the nitrogen mustard is p-[(2-chloroethyl)-(2-mesylethyl)amino] benzoic acid or p-bis(2-mesylethyl)amino-benzoic acid or p-bis(2-trifluoromesylethyl)amino-benzoic acid.

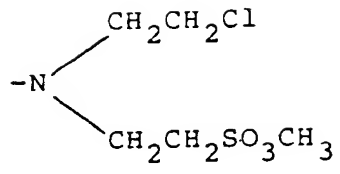
12. A system according to any one of the preceding claims for use in a method of controlling the growth of neoplastic cells in a host involving administration to the host of first components and subsequently, of the second component.

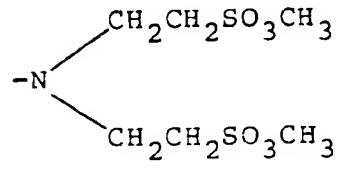
13. A method of controlling the growth of neoplastic cells in a host which comprises administering to the host a first component as defined in any one of claims 1 to 6 followed by administration to the host of a second component as defined in any one of claims 7 to 11.

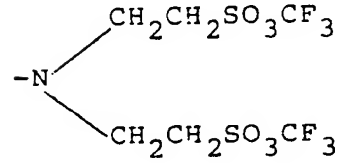
14. A method according to claim 13 wherein each component is administered intravenously.

15. A nitrogen mustard pro-drug of the formula







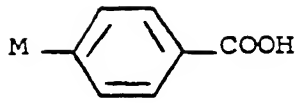
or  group.

16. A pro-drug according to claim 15 wherein R is a glutamic acid residue.

17. A pro-drug according to claim 15 or 16 where R is the residue of a D-amino acid.

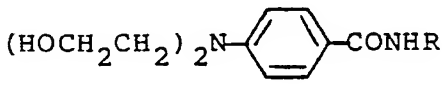
18. A pro-drug according to claim 15 hereinbefore specifically mentioned.

19. A process for preparing a compound as defined in claim 15 which comprises reacting a nitrogen mustard of

formula  or a reactive

carboxy derivative thereof with a carboxy protected amino acid R $NH_2$ and removing the carboxy protecting group.

20. A process for preparing a compound as defined inclaim 15 which comprises reacting a compound of the formula:



with a reagent capable of replacing the HO group by Cl, $CH_3SO_3$ or $CF_3SO_3$.

21. A pharmaceutical composition comprising a pro-drug according to any one of claims 15 to 18 together with a pharmaceutically acceptable carrier or diluent.

22. A composition according to claim 21 suitable for intravenous administration.

23. A conjugate of carboxypeptidase G2 and the $F(ab')_2$ fragment of W14A.

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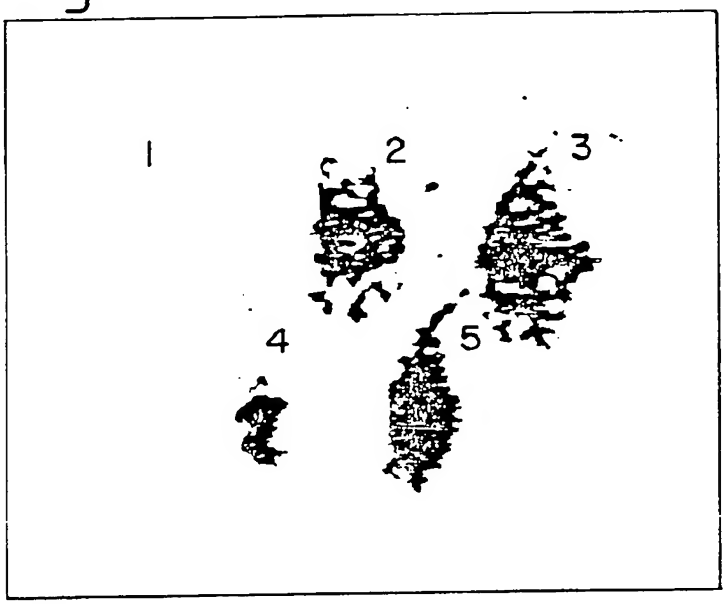
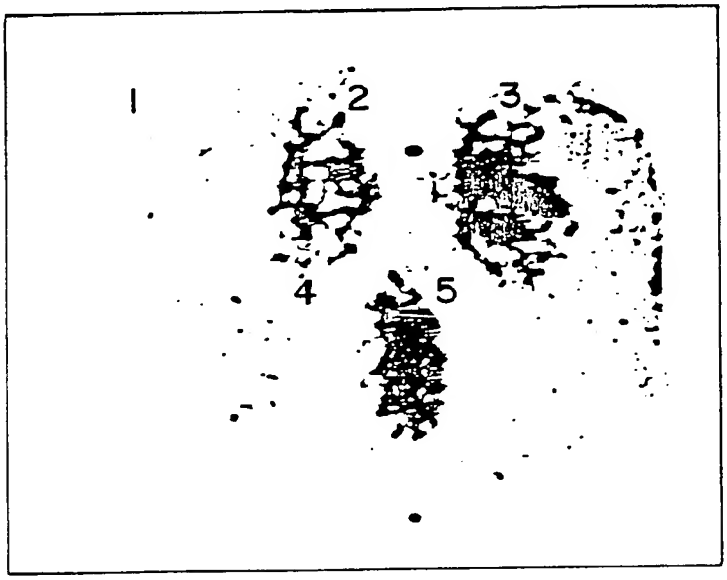
Fig.1.



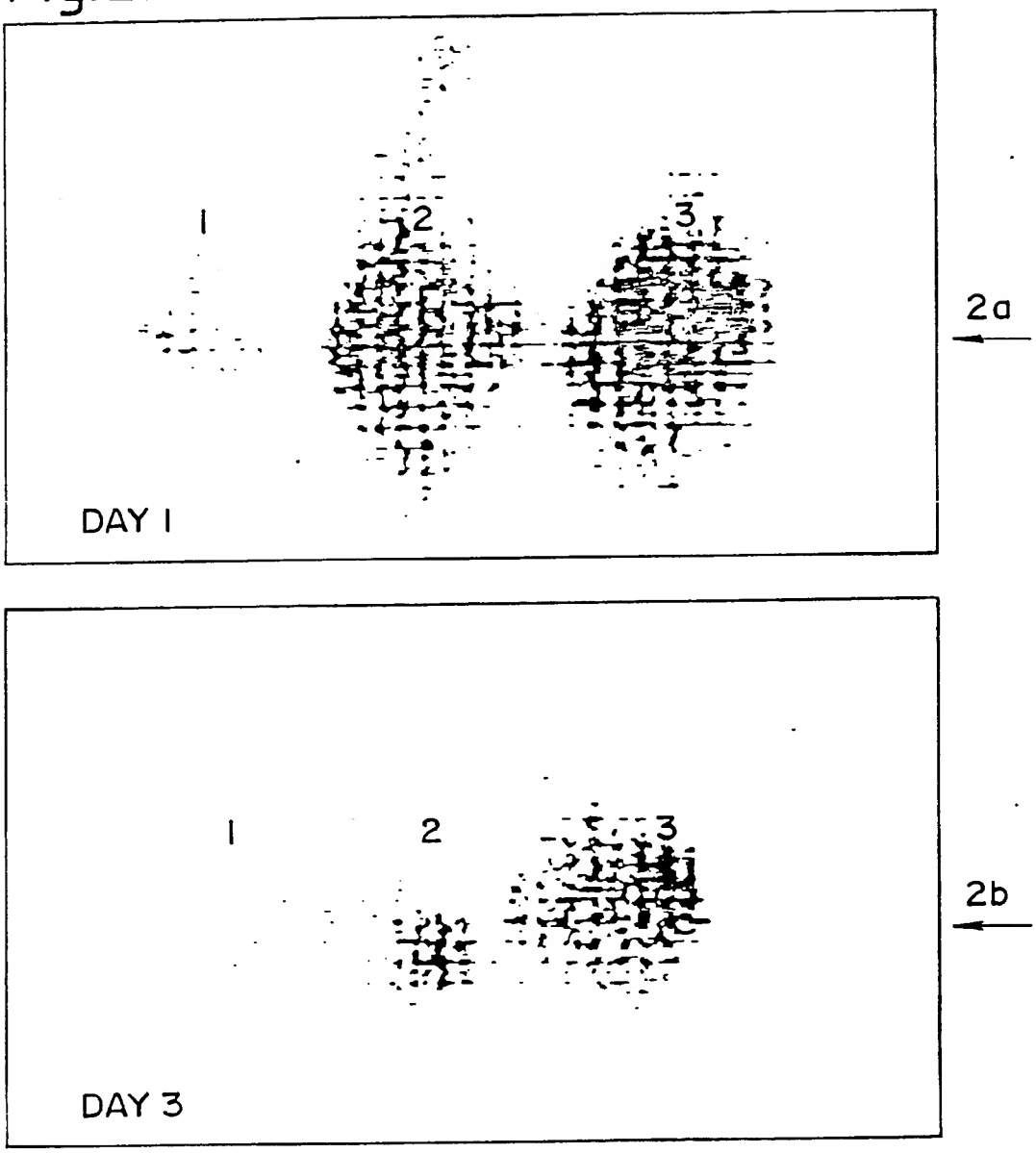




Plates 1a and 1b Nude rats bearing CC3 choriocarcinoma xenografts on their flanks (arrowed) were injected i.v. or i.p. with SPDP- or MBS-linked W14A:CPG2 conjugates; a fifth tumour-bearing rat control was injected with native $^{131}$I-labelled

4. W14A:CPG2 (SPDP-linked) 15.3μCi, 63μg protein, i.p.

5. W14A:CPG2 (MBS-linked) 16.5uCi, 77ug protein, i.p.

Fig.2.




Plates 2a and 2b Nude rats bearing CC3 choriocarcinoma xenografts were injected i.v. with SPDP- or MBS-linked F(ab')$_2$:CPG2 conjugates; a third tumour-bearing rat control was injected with native $^{131}$I-labelled CPG2. 1. Native CPG2, 43uCi, 4lug protein, i.v. 2. F(ab')$_2$:CPG2 (SPDP-linked)

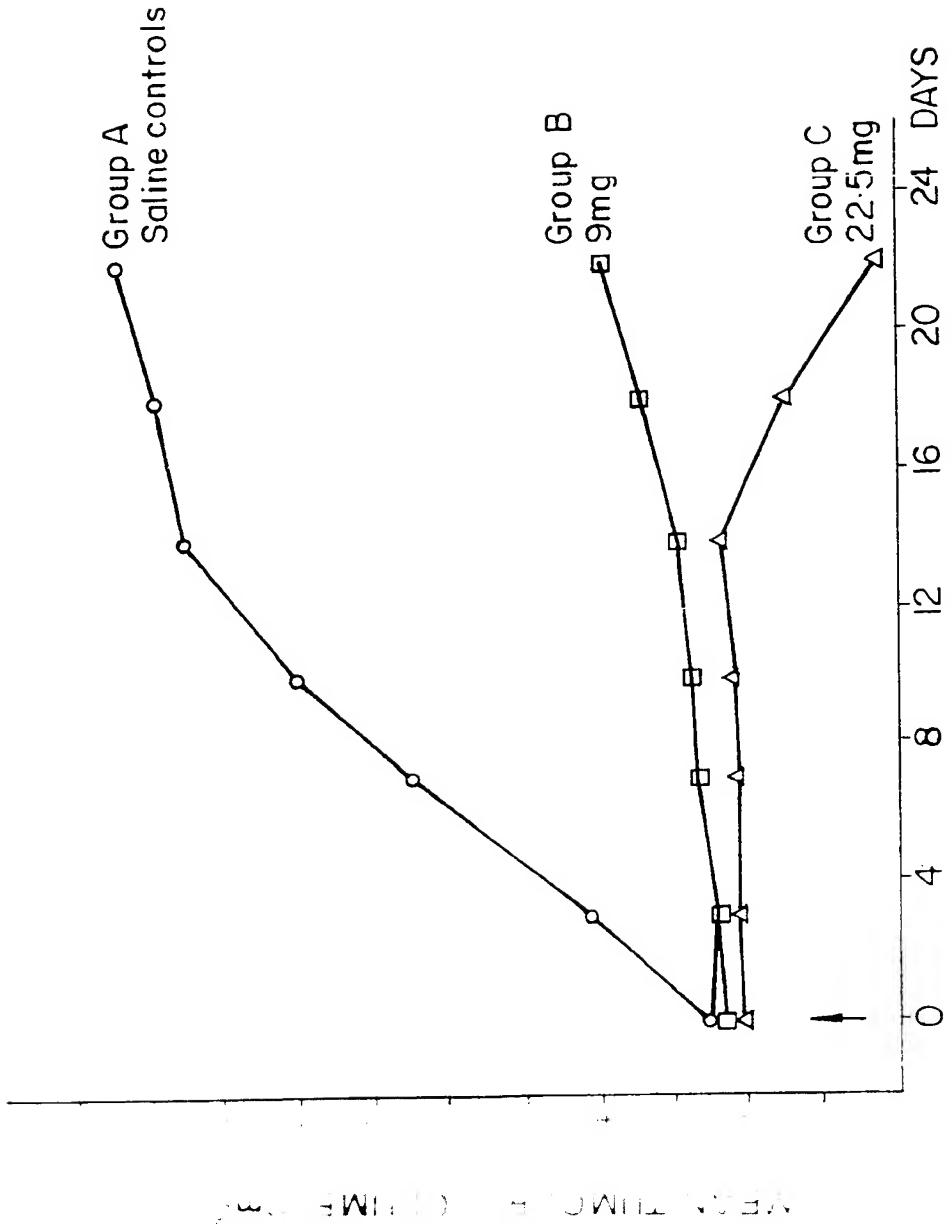



Fig. 3. 'Therapeutic' experiment.

# INTERNATIONAL SEARCH REPORT

International Application No PCT/GB 88/00181

## I. CLASSIFICATION OF SUBJECT MATTER (if several classification symbols apply, indicate all) [6]

According to International Patent Classification (IPC) or to both National Classification and IPC

IPC[4]: A 61 K 39/395; A 61 K 45/05; //(A 61 K 39/395, 31:165)

## II. FIELDS SEARCHED

Minimum Documentation Searched [7]

| Classification System | Classification Symbols |
|---|---|
| IPC[4] | A 61 K |

Documentation Searched other than Minimum Documentation to the Extent that such Documents are Included in the Fields Searched [8]

## III. DOCUMENTS CONSIDERED TO BE RELEVANT [9]

| Category * | Citation of Document, [11] with indication, where appropriate, of the relevant passages [12] | Relevant to Claim No. [13] |
|---|---|---|
| X | FR, A, 2584294 (P. BERDAL) 9 January 1987 see claims 1,2,15 -- | 1,2,5=7 |
| X | Dialog 05464842, 85080842 Medline, A.R. Ahmed et al.: "Cyclophosphamide (Cytoxan). A review on relevant pharmacology and clinical uses", see abstract & J. Am. Acad. Dermatol., Dec 1984, 11(6) p1115-26 -------- | 1,7 |

* Special categories of cited documents: [10]

"A" document defining the general state of the art which is not considered to be of particular relevance

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"P" document published prior to the international filing date but later than the priority date claimed

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"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

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## CERTIFICATION

Date of the Actual Completion of the International Search

8th August 1988

International Searching Authority

EUROPEAN PATENT OFFICE

Signature of Authorized Officer

P.C.G. VAN DER PUTTEN

# ANNEX TO THE INTERNATIONAL SEARCH REPORT ON INTERNATIONAL PATENT APPLICATION NO. GB 8800181 SA 21051

This annex lists the patent family members relating to the patent documents cited in the above-mentioned international search report. The members are as contained in the European Patent Office EDP file on 26/08/88
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| Patent document cited in search report | Publication date | Patent family member(s) | Publication date |
|---|---|---|---|
| FR-A- 2584294 | 09-01-87 | None | |